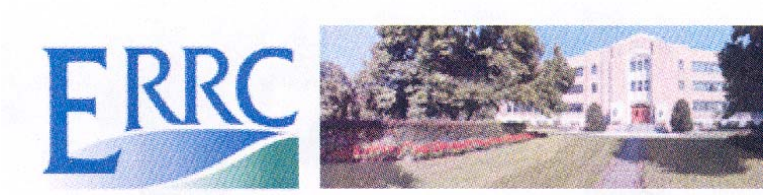

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Title: Effect of Citric Acid on the Radiation Resistance of *Listeria Monocytogenes* and Frankfurter Quality Factors

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Citation: Meat Science (2003) 63: 407-415

Number: 7154

Effect of citric acid on the radiation resistance of *Listeria monocytogenes* and frankfurter quality factors☆

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Received 12 December 2001; received in revised form and accepted 18 April 2002

Abstract

Listeria monocytogenes is a common contaminant of ready-to-eat meat products, including frankfurters. Ionizing (gamma) radiation can eliminate *L. monocytogenes* from frankfurters. Citric acid (CA) is an antioxidant synergist and anti-microbial agent that can be applied to the surfaces of cured meat products prior to packaging. The effect of CA on the radiation resistance of *L. monocytogenes* that was surface-inoculated onto frankfurters was determined. The D_{10} values, the radiation doses required to inactivate 90% of viable *L. monocytogenes*, were 0.61, 0.60, 0.54, and 0.53 kGy, on frankfurters dipped in 0, 1, 5 or 10% CA solution, respectively. CA, although an antioxidant synergist, did not increase antioxidant activity (AA) on frankfurter surfaces as determined by the ferric reducing antioxidant power (FRAP) assay. Lipid oxidation, as determined by the Thiobarbituric acid reactive substances (TBARS) assay, was not affected by CA or ionizing radiation. Color of frankfurters, determined by Hunter *L*, *a*, *b*, indicated that ionizing radiation induced a small, but visually imperceptible, loss of redness (*a*-value). Frankfurter firmness, as measured by maximum shear force, was not affected by ionizing radiation or CA. CA enhanced the lethality of ionizing radiation without negatively impacting frankfurter color, lipid oxidation, firmness, or antioxidant activity.

Keywords: *Listeria monocytogenes*; Ionizing radiation; Gamma irradiation; Citric acid; Frankfurters; Ready-to-eat meat products

1. Introduction

Listeria monocytogenes is a food-borne pathogen capable of growth at refrigerated temperatures and in high salt environments (Smith, 1996). It is a frequent post-process contaminant of ready-to-eat meat products, including frankfurters (Nickelson & Schmidt, 1999). A number of food-borne illness outbreaks and recalls have been attributed to *L. monocytogenes* [Centres for Disease Control (CDC), 1989; MMWR, 1998]. Because of the high mortality rate associated with listeriosis, approximately 20%, *L. monocytogenes* has been given zero tolerance in ready-to-eat meat products [Mead et al., 1999; US Department of Agriculture (USDA), 1989].

Ionizing radiation can eliminate *L. monocytogenes* from raw, cooked, and cured ready-to-eat meat (RTE) products (Monk, Clavero, Beuchat, Doyle, & Brackett, 1995; Thayer, Boyd, Fox, Lakritz, & Hampson, 1995; Thayer, Boyd, Kim, Fox, & Ferrel, 1998). Sommers and Thayer (2000) found that the D_{10} values for *L. monocytogenes*, the dose of ionizing radiation required to eliminate 90% of the microorganism, that were surface-inoculated onto different types of commercial frankfurters ranged from 0.49 to 0.71 kGy, with a mean value of 0.61 kGy. They speculated that product formulation and surface treatments were responsible for those differences.

Citric acid (CA) solution may be applied to the surfaces of cured meats in concentrations up to 10% immediately prior to packaging (Code of Federal Regulations, 1998). Organic acids, including citric acid, exhibit both bacteriocidal and bactiostatic effects against *L. monocytogenes* (Bal'A & Marshall, 1998; Buchanan & Golden, 1994, 1998; Ita & Hutkins, 1991). The antilisterial activity of citric acid is dependent on pH, concentration and anion effects (Young & Foegeding,

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0309-1740/02/$ - see front matter Published by Elsevier Science Ltd.
PII: S0309-1740(02)00100-6

1993). Some studies suggest that organic acids may enhance the bacteriocidal effects of ionizing radiation (Bhide, Parturkar, Sherikar & Waskar, 2001; Giroux et al., 2001). Relatively little, if any, data exist on the combined effects of ionizing radiation and citric acid on the survival of *L. monocytogenes* on RTE meats.

This work addressed following questions: (1) To what extent does CA affect the viability of *L. monocytogenes* surface-inoculated onto frankfurter surfaces? (2) Does application of a citric acid solution to the surface of frankfurters affect the radiation resistance of *L. monocytogenes*? and (3) Does application of citric acid solution to frankfurter surfaces, in combination with ionizing radiation, affect frankfurter quality factors including surface antioxidant power, lipid oxidation, color, and shear force?

2. Materials and methods

2.1. Strains

Four *L. monocytogenes* strains (H7762, H7596, H7769 and H7969) isolated from RTE meats following *Listeriosis* outbreaks were obtained from the Centers for Disease Control and Prevention (Atlanta, GA). The strains were propagated on Palcam Medium (Difco Laboratories, Detroit, MI) at 37 °C and maintained at 0–2 °C until ready for use. Strain identity was confirmed by Gram Stain followed by analysis with Gram Positive Identification (GPI) cards using the Vitek Automicrobic System (bioMerieux Vitek, Inc., Hazelwood MO).

2.2. Bacterial cultures

Each *L. monocytogenes* strain was cultured independently in 100 ml Tryptic Soy Broth (Difco Laboratories, Detroit, MI) in a baffled 500 ml Erlenmeyer culture flask at 37 °C (150 rpm) for 18 h. The cultures were then combined and the mixture pelleted by centrifugation (1725 × *g*). The *L. monocytogenes* cells were then concentrated 10-fold by re-suspension in 40 ml of Butterfield's Phosphate Buffer (BPB) (Applied Research Institute, Newtown, CT).

2.3. Citric acid dip, inoculation and packaging

Mixed meat (beef and pork) frankfurters from the same batch were obtained from a local manufacturer and stored at −20 °C until ready for use. Frankfurters were then thawed and dipped in sterile deionized water or filter sterilized 1, 5, or 10% (w/v) citric acid monohydrate solution. Following dipping the frankfurters were organized, four per pack, into Fresh-Pack gas impermeable bags. Each frankfurter pack was then inoculated with 200 μl of *L. monocytogenes* cocktail. The frankfurters were vacuum-packed to 0.3 mmHg using a Multi-Vac Model A300 packager (Multi-Vac, Kansas City MO) and stored at 0–2 °C until irradiation (approximately 30 min).

2.4. Gamma irradiation

A Lockheed Georgia Company (Marietta, GA) self-contained ^{137}Cs gamma irradiation source was used. The radiation source consisted of 23 individually sealed source pencils placed in an annular array. The 22.9×63.5 cm cylindrical sample chamber was located central to the array when placed in the operating position. The inoculated vacuum-packaged frankfurters were placed centrally in the sample chamber to insure uniformity of dose.

The dose rate provided by the irradiator was 0.098 kGy/min. The temperature during irradiation process was maintained at 4.0(±1.0) °C by the gas phase of a liquid nitrogen source which was introduced directly into the top of the sample chamber. The temperature was monitored using two thermocouples placed 1–2 cm adjacent to the samples. The dose delivered was verified by use of alanine pellet dosimeters, which were then measured using a Bruker EMS 104 EPR Analyzer. The radiation doses were 0.4, 0.8, 1.2, 1.6, 2.0 and 2.4 kGy.

2.5. D_{10} value

D_{10} value is defined as the radiation dose required to produce a 90% reduction in viable organism. The average (N) CFU/cm^2 of an irradiated sample was divided by the average CFU/cm^2 of the untreated control (N_o) to produce a survivor ratio (N/N_o). The D_{10} value was then determined by calculating the reciprocal of the slope provided by the (N/N_o) ratios (Ley, 1983). The 0.4 through 2.4 kGy doses were used for determination of D_{10} value. Each experiment was conducted independently three times.

Following irradiation frankfurters were rinsed in BPB, the rinse serially diluted, and 1 ml of the appropriate dilutions pour plated on TSA Medium as described by Sommers and Thayer (2000). Background microflora, as determined by plating on TSA using the rinse from uninoculated frankfurters, was less than two $\log_{10}$ CFU/cm^2. This was insufficient to influence D_{10} value determination. The same inoculation, irradiation, and plating procedures were used for the 4 week storage study.

2.6. FRAP assay

Total antioxidant power of cured meat was measured directly by the Ferric Reducing Antioxidant Power (FRAP) assay (Benzie & Strain, 1999). In the assay, the antioxidants present reduce ferric tripyridyltriazine to the ferrous form, which has an intense blue color. Absorbance was measured at 593 nm and concentration

calculated against a standard curve of sodium erythorbate (0–500 μM). Results of the FRAP assay are expressed in terms of μmoles antioxidant per cm^2 of frankfurter surface area as determined using the surface area of a cylinder and sphere halves.

Hot dogs were vacuum-packaged ($n=6$) and irradiated as previously described to doses of 0, 1.2 and 2.4 kGy. Upon cutting open a sample, 40 ml of distilled water was added and the frankfurters shaken with the water for 30 s. The rinse water was added to a 100 ml volumetric flask. A second 40 ml of water was added to the bag and the sample shaken another 30 s and the rinses combined in the flask. The flask was filled to the mark with distilled water and the FRAP value determined using the rinse.

2.7. Lipid oxidation

Lipid oxidation was measured using the thiobarbituric acid (TBA) assay modified from the methods of Hodges, DeLong, Forney, and Prange (1989) and Zipser and Watts (1962). Ten grams of frankfurter were homogenized with 25 ml 0.5 M phosphate (pH 2.5) buffer containing 0.08% sulfanilamide and 0.01% BHT using a homogenizer (Virtishear, Virtis, Gardiner, NY) at a speed setting of 70 for 1 min. The homogenate was filtered through Whatman No. 2 paper filter (Whatman, Clifton, NJ), and then the filtrate was centrifuged at 1300 *g* for 10 min at 5 °C in a Sorvall RT6000B refrigerated centrifuge (DuPout Co., Wilmington, DE). A 1.6 ml supernatant aliquot was added to a test tube containing 1.6 ml of either (i) −TBA solution: 20% (w/v) trichloroacetic acid and 0.01% butylated hydroxytoluene or (ii) +TBA solution: containing the above plus 0.65% TBA. Samples were then mixed vigorously, heated at 95 °C in a water bath for 25 min, cooled and centrifuged at 1300 *g* for 10 min at 5 °C. Absorbances at 440, 532, and 600 nm were monitored using a Shimadzu UV-1601 spectrophotometer (Shimadzu Scientific Instruments, Columbia, MD). TBA reactive substance (TBARS) values are expressed as malondialdehyde (MDA) equivalent and calculated using the formulas developed by Hodges et al. (1989).

$$[(Abs532_{+TBA} - Abs600_{+TBA}) - (Abs532_{-TBA} - Abs600_{-TBA})] = A. \quad (1)$$

$$[(Abs440_{+TBA} - Abs600_{+TBA})0.0571] = B. \quad (2)$$

$$MDA(nmol.g^{-1}) = [(A - B)/157000]10^6. \quad (3)$$

2.8. Color measurement

Frankfurters were vacuum-packaged and irradiated as described above. Frankfurters were cut into approximately 4 cm lengths. The internal portion was removed with a knife and the remaining portion, a rectangular piece approximately 3 mm thick, was flattened onto the view port cover and held in place with a standard Petri plate. Color analysis was then performed using a Hunter Miniscan XE meter (Hunter Laboratory, Inc., Reston, VA) as outlined by Nanke, Sebranek, and Olsen (1998, 1999). The meter was calibrated using black and white standard tiles, Illuminate D65, 10° standard observer, and a 2.5 cm port/viewing area. Six readings were taken per parameter.

2.9. Shear force

Cutting force of hot dogs was performed using a Texture Technologies Corp. (Scarsdale, NY) TA-XT2 Texture Analyzer. A TA-7 Warner-Bratzler Blade was used with a test speed of 2.0 mm/s, 30 mm distance, and 20 g auto-trigger. Maximum shear force (*g*) was performed six times ($n=6$) per parameter.

2.10. Statistical analysis

Statistical analysis was completed using SAS/STAT Version 6.12 (SAS Institute, Inc., Cary, NC) or Sigma Plot Version 6.0 (Chicago, IL). Population reduction data were analyzed by analysis of variance using the general linear model procedure of the SAS statistical package. Comparison of regressions was performed by analysis of covariance (Freund, Little, & Spector, 1986; Sommers & Thayer, 2000).

3. Results

The pH of frankfurter surfaces were approximately 6.5, 6.3, 4.5, and 3.0 for frankfurters dipped in 0, 1, 5 or 10% CA solution, respectively as determined using a microprobe. Surface pH was not affected by irradiation (data not shown). Survival of *L. monocytogenes* surface-inoculated onto frankfurters dipped in 0, 1, 5, and 10% CA solution and incubated for approximately 60 min (0 kGy control) was determined. When *L. monocytogenes* was inoculated onto frankfurter packs viability decreased from 1.7×10^9 CFU on untreated frankfurters to 0.7×10^9 CFU on frankfurters dipped in 10% CA, which was statistically significant as determined by ANOVA ($n=3$, $\alpha=0.05$) (Fig. 1A). When *L. monocytogenes* was surface-inoculated onto frankfurter packs with 80.6×10^3 CFU *L. monocytogenes* viability decreased significantly, to 8.11×10^3 CFU on franks dipped in 10% CA solution as determined by ANOVA ($n=3$, $\alpha=0.05$) (Fig. 1b). Although the decreased survival of *L. monocytogenes* in 5% CA was reproducible between experiments, at both the higher and lower inoculum, levels it was not statistically significant.

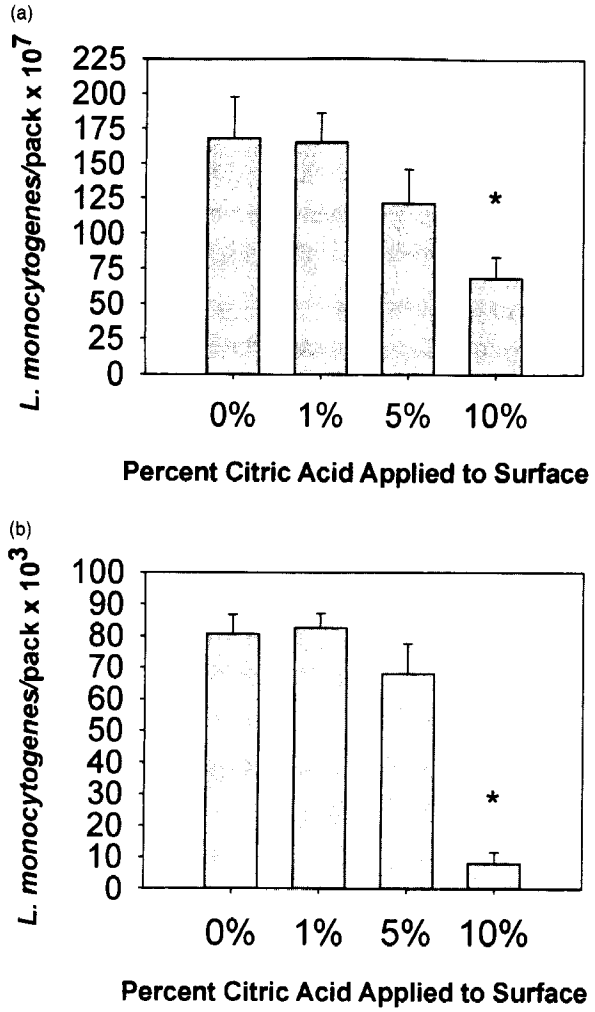

Fig. 1. Survival of *Listeria monocytogenes* on frankfurters prior to irradiation. Frankfurters were dipped in water or 1, 5 or 10% citric acid solution, inoculated with (A) 1.7×10^9 CFU/pack and (B) 8.06×10^4 CFU/pack *L. monocytogenes* cocktail and vacuum-packaged. Survival was assessed following a 30 min incubation at 4 °C. Each experiment was conducted independently three times ($n = 3$). Standard error bars are shown for each parameter tested. *Significant decrease in D_{10} value versus the untreated control as determined by ANOVA ($n = 3$, $\alpha = 0.05$).

The radiation resistance, D_{10} values of *L. monocytogenes* surface-inoculated into frankfurter packs with and without CA solutions, was determined (Fig. 2). D_{10} values of 0.60, 0.65, 0.54, and 0.53 kGy were obtained for *L. monocytogenes* surface-inoculated onto frankfurters dipped in 0, 1, 5, and 10% CA, respectively. The total $\log_{10}$ reduction observed for irradiated *L. monocytogenes* in the presence of 10% CA, at all radiation doses, as opposed to those irradiated in the presence of 1 or 5% CA, was considerable. This was due to selective killing of radiation damaged *L. monocytogenes* at the higher acid concentration (data not shown). When compared using ANCOVA ($n = 3$, $\alpha = 0.05$) the D_{10} values for *L. monocytogenes* on the 5% and 10% CA dipped frankfurters were significantly less than those for the 0% CA control.

Citric acid, which can applied to the product surfaces immediately prior to packaging, is used as an antioxidant synergist in the production of cured meat products [Code of Federal Regulations (CFR), 1998]. The antioxidant power of citric acid, using sodium erythorbate as a reference antioxidant, was determined (Fig. 3) using the Ferric Reducing Antioxidant Power (FRAP) assay. At 500 μM the absorbance (593 nm) was 0.713 for sodium erythorbate versus 0.011 for CA (Fig. 3A). Citric acid was a relatively week antioxidant in comparison to sodium erythorbate.

When the FRAP assay was used to determine surface antioxidant power of frankfurters dipped in 0, 1, 5, and 10% CA solutions the FRAP values ranged from 23.6 μM/cm^2 (0% CA) to 17.6 μM/cm^2 for frankfurters dipped in 10% CA (Fig. 3B). Variability in the data is due to the fact that the FRAP values obtained were close to the lower detection limit for the assay on RTE meats. FRAP values were not affected by exposure to ionizing radiation doses up to 2.4 kGy (data not shown).

The effect of CA and ionizing radiation on lipid oxidation was measured using the TBARS assay (Table 1). Lipid oxidation, expressed as μg malondialdehyde/g of frankfurter ranged from a low of 1.51 μg/g to a high of 2.08 μg/g in the samples tested. Ionizing radiation (up to 2.4 kGy) did not affect lipid oxidation in the non-acid dipped control frankfurters. Lipid oxidation was reduced in unirradiated CA dipped franks in comparison to the unirradiated non-acid dipped frankfurters (Table 1) as determined by ANOVA ($n = 3$, $\alpha = 0.05$). Lipid oxidation increased in irradiated CA dipped frankfurters ($n = 3$, $\alpha = 0.05$). However, the TBARS values obtained were less than those obtained from non-acid dipped frankfurters (Table 1).

Ionizing radiation can affect the color of raw meat and cured meat products. Although visually imperceptible to the authors, ionizing radiation produced statistically significant decreases in frankfurter redness (*a*-value), yellowness (*b*-value) and *L*-value (brightness) as determined by ANOVA ($n = 5$, $\alpha = 0.05$) (Table 1). The application of CA did not affect *a*-value or *b*-value, but significantly reduced *L*-value as determined by ANOVA ($n = 5$, $\alpha = 0.05$) (Table 1). However, the application of both CA and ionizing radiation positively impacted both *b*-value and *L*-value as determined by ANOVA ($n = 5$, $\alpha = 0.05$) (Table 1). Shear force (firmness) of CA dipped and irradiated frankfurters, was determined. Neither CA, up to 10%, or ionizing radiation, up to 2.4 kGy, affected maximum shear force (Table 1).

The ability of *L. monocytogenes* to grow on CA dipped and vacuum-packaged irradiated (10^9 *L. monocytogenes* per four frankfurter pack and then 2.4 kGy) and unirradiated (10^4 *L. monocytogenes* per four frankfurter pack and 0 kGy) was determined (Fig. 4). The frankfurters were stored for 4 weeks at 9 °C. *Listeria* specific counts were determined by plating on Palcam

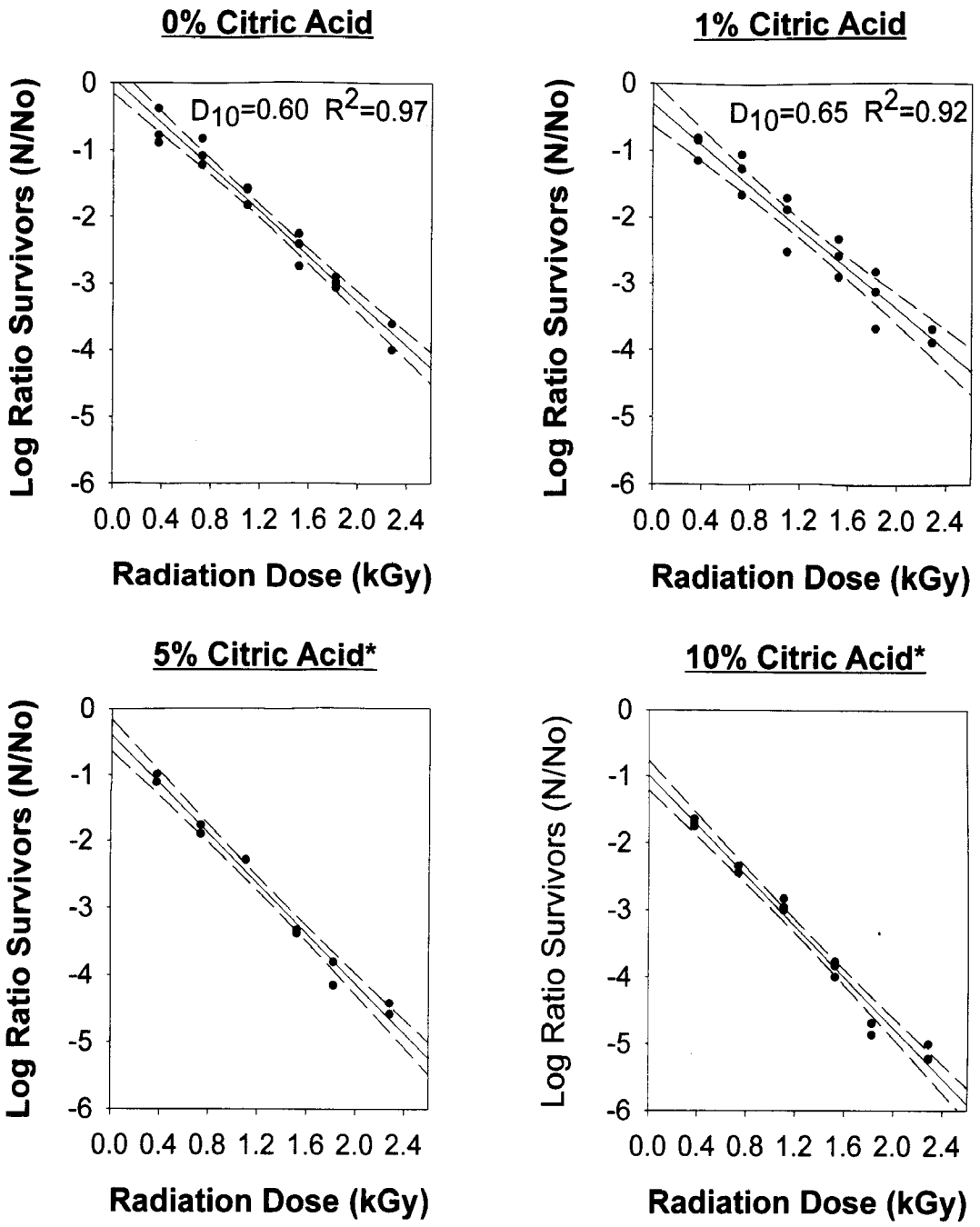

Fig. 2. Radiation resistance of *Listeria monocytogenes* surface-inoculated onto frankfurters in the presence or absence of citric acid. Each experiment was conducted independently three times ($n=3$), except for 5% citric acid, which was conducted twice ($n=2$). Individual $\log_{10}$ reduction data points are shown as closed circles. The regression is shown as a solid line. Confidence intervals (95%) are shown as dashed lines. *Significant decrease in D_{10} value versus the untreated control as determined by ANCOVA ($n=3$, $\alpha=0.05$).

medium while non-specific counts were determined by plating on Tryptic Soy agar. As shown, both unirradiated and irradiated *L. monocytogenes* and background microflora were capable of growth on frankfurters dipped in sterile water. However, *L. monocytogenes* was unable to grow on either the irradiated or unirradiated frankfurters dipped in 5 or 10% CA. In contrast, the background microflora, primarily lactic acid bacilli as determined by plating on TSA and MRS agar, was able to proliferate regardless of treatment with CA or ionizing radiation.

4. Discussion

Organic acids can inhibit microbial growth and are routinely used as preservatives. Organic acids, including citric acid, have been tested in solution studies for their

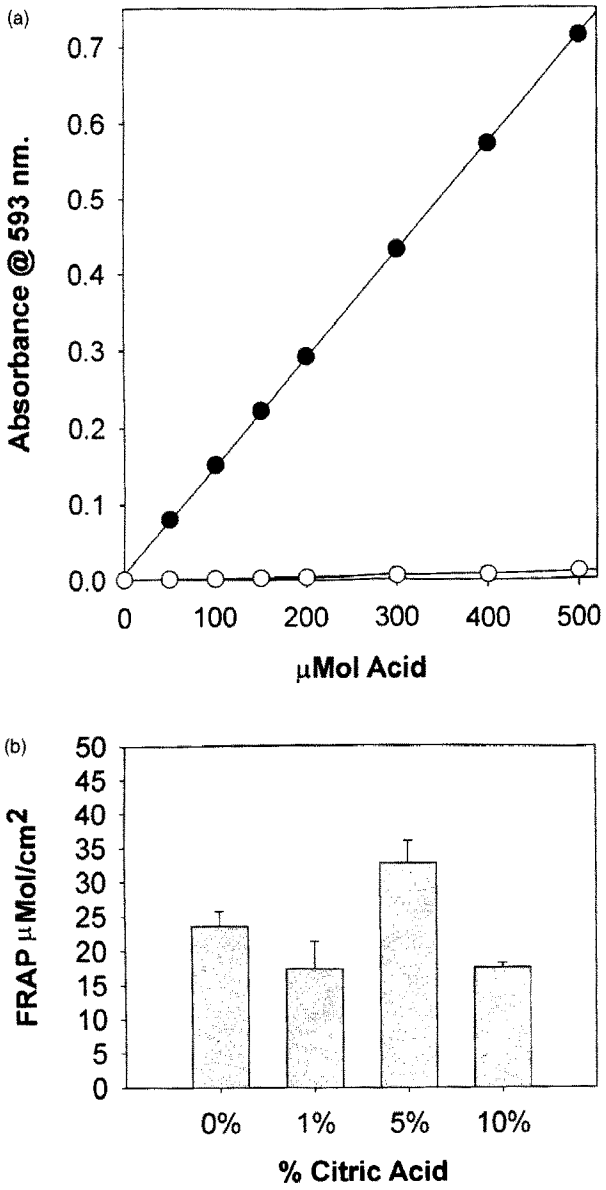

Fig. 3. Ferric antioxidant power (FRAP) assay. (A) Absorbance (593 nm) of citric acid versus sodium erythorbate. (B) FRAP values for frankfurter surfaces dipped in water, 1, 5 and 10% citric acids solution expressed as µM antioxidant/cm^2 surface area. Each experiment was conducted independently three times ($n=3$). Standard error bars are shown for each parameter tested.

antilisterial activity (Bal'A & Marshall, 1998; Buchanan & Golden, 1994, 1998; Ita & Hutkins, 1991; Young & Foegeding, 1993). CA was used in this study because of the high concentrations that can be applied to the surfaces of cured meat products. CA, an antimicrobial and antioxidant synergist, has been approved for use as a food additive and can be used in concentrations up to 10% for application to the surfaces of cured meat products (CFR, 1998). Both ionizing radiation and citric acid exhibit bacteriocidal effects against *L. monocytogenes* on RTE meats (Palumbo & Williams, 1994; Sommers & Thayer, 2000).

The effect of citric acid, in combination with ionizing irradiation, on the survival of *L. monocytogenes* was determined. The D_{10} value of *L. monocytogenes* surface-inoculated onto water-dipped frankfurters was 0.60 kGy, which was consistent with the value previously obtained by Sommers and Thayer (2000). Sommers, Handel, Fan, and Niemira (in press) found that the antioxidants in soy protein concentrate, at concentrations commonly used in RTE meats, could increase the radiation resistance of *L. monocytogenes*. In this study, the weak antioxidant citric acid sensitized, as opposed to protected, *L. monocytogenes* against the lethal effects of ionizing radiation.

As with many food processing technologies excessive treatment has the potential to adversely affect product quality factors. For instance, ionizing radiation has the potential to induce both lipid oxidation, color changes, and texture changes in raw and RTE meats (Ahn et al., 1998; Chen, Jo, Lee, & Ahn, 1999; Kamat, Paul, D'Souza, & Thomas, 1997; Nanke et al., 1998, 1999). Although statistically significant differences were observed in lipid oxidation in CA treated and irradiated frankfurters the values did not exceed those obtained from the control frankfurters. CA and ionizing radiation had little affect on AA as determined by FRAP assay. The values obtained were at the lower limit of detection for the assay.

Although statistically significantly changes were observed in frankfurter color as a result of both CA treatment and exposure to ionizing radiation, those changes were minimal and were detectable only by instrument. Frankfurter shear force (firmness) was not affected by either CA or by treatment with ionizing radiation. This is consistent with previous work conducted on irradiated, but not acid dipped, frankfurters (Barbut, Maurer, & Thayer, 1988; Terrell, Heiligman, Smith, Wierbicki, & Carpenter, 1981, Terrell, Smith, Heiligman, & Carpenter, 1981; Terrell, Swasdee, Smith, Heiligman, & Carpenter, 1981). Overall, CA and ionizing radiation had little effect on the quality factors tested at the radiation doses used. It may be possible to use combination treatments such as organic acids and ionizing radiation to minimize negative impacts on quality factors that would be observed with individual treatments alone.

Future work should include the effect of other organic acids on the radiation resistance of *L. monocytogenes* on cured RTE meat products. Buchanan and Golden (1994) indicated that citric acid exerted its antimicrobial effects via pH dependent and independent mechanisms. Wilson, Curry, Unklesby, Ionnotti, and Unklesby (1999) found that lactic acid incorporated into ground beef had no effect on the radiation resistance of *E. coli* O157:H7 while Giroux et al. (2001) found the opposite for other organic acids. Sommers and Niemira (unpublished data) found the acidulants sodium diacetate and

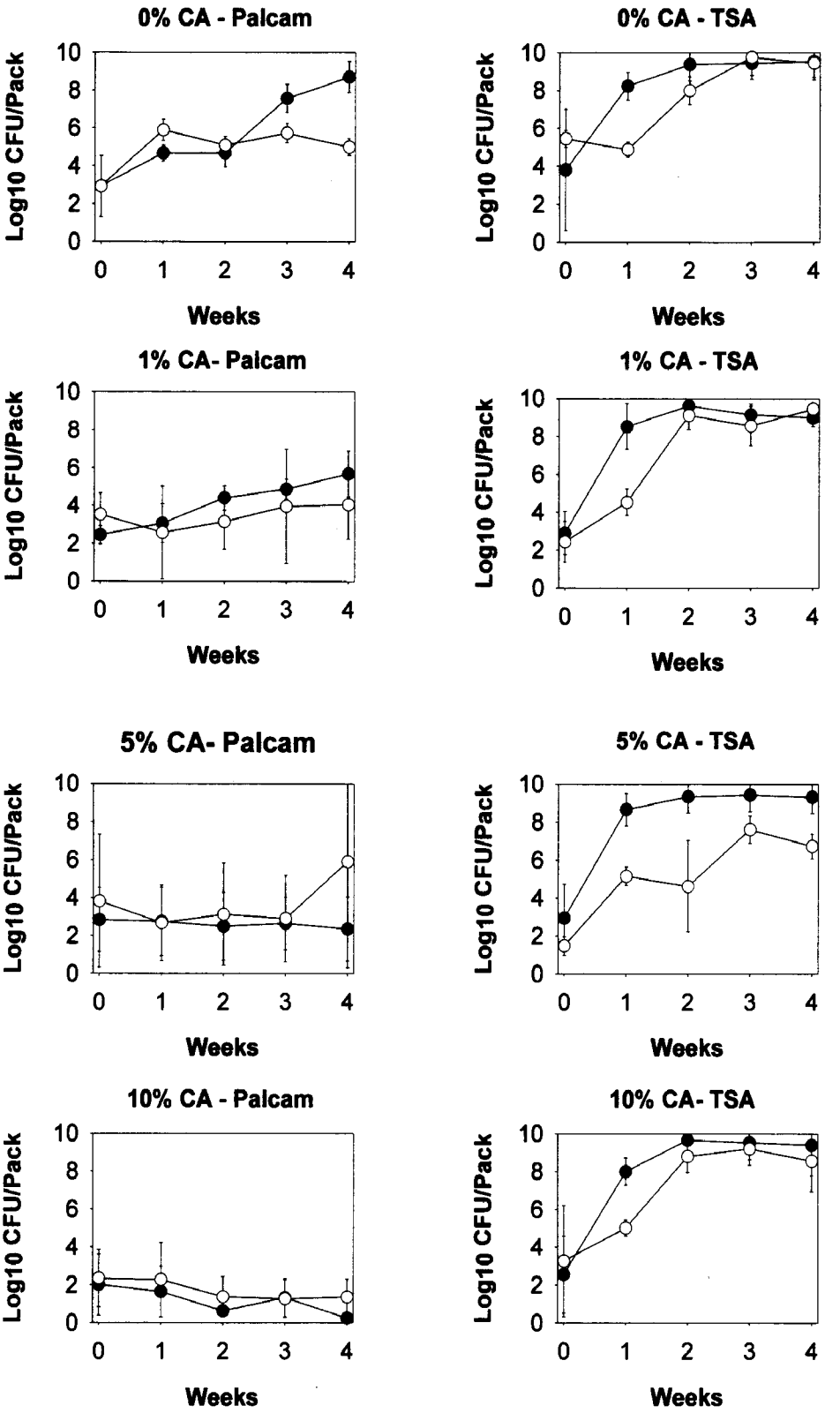

Fig. 4. Growth of *Listeria monocytogenes* and background microflora on frankfurters dipped in 0, 5 or 10% citric acid solution during refrigerated storage (9 °C). *Listeria* specific counts were determined using Palcam Medium. Total aerobic plate counts (APC's) were determined using tryptic soy agar. Closed circles were unirradiated samples and open circles were irradiated samples. Each experiment was conducted independently three times ($n = 3$). Standard error bars are shown for each parameter tested.

Table 1
Color, lipid oxidation and firmness of frankfurters dipped in citric acid and treated with ionizing radiation

	Radiation dose (kGy)	*a*-Value (redness)	*b*-Value (yellowness)	*L*-value (brightness)	TBARS (μg/g MDA)	Shear force (*g*)
0% Citric acid	0	16.7 (±0.21)a	18.6 (±0.32)a	46.6 (±0.20)a	1.95 (±0.04)a	1238 (±93.4)a
	1.2	16.3 (±0.21)a	18.3 (±0.17)a	45.9 (±0.24)a	1.86 (±0.09)a	1334 (±68.4)a
	2.4	15.5 (±0.37)b	17.9 (±0.10)b	45.8 (±0.20)a	1.85 (±0.21)a	1242 (±53.2)a
1% Citric acid	0	17.0 (±0.25)a	18.3 (±0.10)a	45.7 (±0.29)a	1.67 (±0.18)b	1358 (±49.4)a
	1.2	16.1 (±0.26)b	17.6 (±0.21)b	45.8 (±0.37)a	1.86 (±0.09)a	1325 (±28.7)a
	2.4	15.6 (±0.24)b	18.2 (±0.15)a	45.5 (±0.35)b	2.01 (±0.11)a	1322 (±76.9)a
5% Citric acid	0	16.9 (±0.15)a	18.3 (±0.10)a	45.7 (±0.29)a	1.56 (±0.14)b	1523 (±23.6)b
	1.2	16.1 (±0.26)b	17.6 (±0.21)b	45.4 (±0.25)a	1.84 (±0.07)a	1432 (±56.8)a
	2.4	16.5 (±0.12)a	18.2 (±0.10)a	45.4 (±0.17)b	1.93 (±0.11)a	1366 (±96.3)a
10% Citric acid	0	15.8 (±0.19)b	18.1 (±0.15)a	45.3 (±0.19)b	1.71 (±0.07)b	1376 (±92.2)a
	1.2	15.6 (±0.31)b	18.0 (±0.22)a	46.1 (±0.25)a	1.86 (±0.06)a	1407 (±42.3)a
	2.4	16.5 (±0.27)a	18.4 (±0.10)a	45.9 (±0.28)a	2.08 (±0.07)a	1346 (±72.5)a

L-values (brightness), *a*-values (redness) and *b*-values (yellowness) are shown for each parameter tested ($n = 6$). Lipid oxidation was determined using the thiobarbituric acid reacting substances (TBARS) assay and expressed as μg of malondialdehyde (MDA) per gram of meat ($n = 3$). Firmness was measured by shear force ($n = 5$). Standard errors of the means are shown in parenthesis. Statistically significant differences are indicated by letter within each column ($\alpha = 0.05$).

hydrochloric acid (pH 4.5) had no effect on the radiation resistance of *L. monocytogenes* when inoculated onto frankfurter emulsion while glucono-delta-lactone increased the radiation sensitivity of *L. monocytogenes* suspended in tofu.

Other combination treatments might include the use of ionizing radiation and in-package surface heat treatment of RTE meats. Microorganisms, including *L. monocytogenes*, are more sensitive to heat following sub-lethal doses of ionizing radiation (Grant & Patterson, 1998). In addition, Juneja and Eblen (1999) determined that *L. monocytogenes* was more sensitive to heat in the presence of organic acid. A realistic possibility for elimination of *L. monocytogenes* from RTE meat products that are sensitive to quality changes might include surface treatment with organic acid prior to packaging, followed by irradiation and a short in-package surface heat-treatment.

Acknowledgements

We would like to thank L. Melenski, N. Keser and R. Richardson for technical assistance.

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